

A FOURTH VARIETY OF TRICHONOCARDIASIS, WITH A NOTE ON THE CULTIVATION OF *NOCARDIA TENUIS*
(CASTELLANI, 1911)

BY

J. W. SCOTT MACFIE, D.Sc., M.B.

WEST AFRICAN MEDICAL STAFF

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INTRODUCTION

Trichonocardiasis is by no means uncommon in West Africa, but, so far as I am aware, its occurrence in this part of Africa has not previously been recorded, with the exception of a brief reference to Trichonocardiasis flava in Europeans on the Gold Coast contained in the excellent account of the occurrence of this condition in the Sudan published by Chalmers and O'Farrell (1913). In its introduction this paper was claimed to be the first record of the occurrence of Trichonocardiasis in Africa, but further on (p. 527) mention was made of 'a condition resembling Trichonocardiasis flava in Europeans on the Gold Coast, West Africa,' observed by one of the authors as long ago as 1898. As Castellani did not describe the disease until 1911 the diagnosis of the condition observed in 1898 was presumably retrospective, and it is perhaps of some interest to confirm the record.

This disease, which affects the shafts of the hairs in the axilla and groin, is caused by a fungus *Nocardia tenuis*, either alone or associated with a chromogenic organism. Three varieties are recognised, namely, Trichonocardiasis flava in which only the fungus is present, Trichonocardiasis nigra in which it is associated with *Micrococcus nigrescens*, and Trichonocardiasis rubra in which it is associated with *Micrococcus castellanii*. A fourth variety, which from its brownish colour might appropriately be called Trichonocardiasis fusca, occurs on the Gold Coast, and a short description of this form is given in this paper.

TRICHONOCARDIASIS FUSCA

In a case of Trichonocardiasis examined recently the nodules on the hairs were of a brownish or light brick-red colour, quite distinct from the yellow tint of Trichonocardiasis flava or the rich red colour of Trichonocardiasis rubra.

Dr. A. Ingram, who very kindly sent me the materials from this case and to whom I have much pleasure in acknowledging my thanks, stated that the patient was a European who had been aware of the presence of the affection for the last three years. When the patient was in England the nodules disappeared, but always reappeared a month or two after his return to West Africa. The hairs affected were those of the axillae.

Scrapings from the hairs, when sown on ordinary agar or ascitic agar, produced an abundant crop of yellow colonies. The organism itself was colourless, non-motile, and had the characteristic appearance of a diplococcus. It stained well with all the ordinary reagents, but when treated by Gram's method the result was not uniform: some of the cocci showed a positive reaction, others a negative, and not infrequently one segment of a diplococcus was seen to have retained the stain while the other had been completely decolourised. The same peculiarity was observed by Chalmers and O'Farrell in the case of *Micrococcus castellanii*.

On agar, glucose agar, and ascitic agar the diplococcus grew well, producing yellow colonies of a shade similar to that depicted by Chalmers and O'Farrell as typical of young cultures of *M. castellanii*, and which they describe as being lemon-chrome coloured. In this case, however, the colonies never developed a red colour, no matter how old they were nor on what medium they were growing; indeed, in very old cultures the yellow colour became if anything slightly paler. In nutrient broth an abundant growth took place, producing a general turbidity and throwing down a white precipitate, but the yellow colour was not obvious in this medium. No indol was produced in peptone water, and no fluorescence in neutral red medium. Neither acid nor gas was produced after a week's cultivation in the following sugary and alcoholic peptone media: glucose, laevulose, galactose, maltose, lactose, saccharose, dulcitate, and mannite. Unfortunately it was

impossible to test these reactions further owing to a lack of the rarer carbohydrates, &c., which are at present unobtainable.

So far as it was possible to test its properties, this organism seemed to resemble in its general characters the cocci associated with the red and black varieties of trichonocardiasis, but differed from them in the colour of the pigment it produced.

OTHER FORMS OF TRICHONOCARDIASIS

Europeans on the Gold Coast are not infrequently affected by trichonocardiasis, but as the disease causes little or no inconvenience, and as it usually attacks the hairs of the axillae which are not always easy to scrutinize, it is often overlooked by the patient himself. It is probable, therefore, that the condition is much more common than is generally supposed.

Up to the present I have observed, in addition to the variety described above, the two forms known as *Trichonocardiasis flava* and *Trichonocardiasis rubra*. From the latter variety a diplococcus has been isolated which conforms morphologically and culturally to the description of *Micrococcus castellanii* given by Chalmers and O'Farrell. I have not yet seen a case of the black variety, *Trichonocardiasis nigra*.

With regard to the natives, it is difficult to determine to what extent they are subject to the disease since most of the races shave the axillae. Chalmers and O'Farrell suggest that this practice may have originated, for one reason, because the occurrence of trichonocardiasis had been observed by the native doctors. If this was so, the fact seems to have been forgotten, for now-a-days a native, if asked why he shaves the axillae, will generally reply that it is because it prevents excessive perspiration.

The pathology and microscopical anatomy of this affection have been fully described by Chalmers and O'Farrell. Only the shafts of the hairs appear to be affected, and the nodules when discovered by accident, as they generally are, are often mistaken for dirt or dried sweat until it is found that no amount of washing will remove them. It has been stated that the hairs are not affected by the growth, but of this I am uncertain, as I have observed that the diseased hairs

in Europeans appeared to be discoloured or greyer than the healthy ones, and Dr. Ingram, in forwarding to me the materials from the case of *Trichonocardiasis fusca*, stated that in this patient the hairs in the right axilla, which were heavily infected, were nearly all grey, whereas those in the left axilla, which were but little affected, retained their pigment.

The infection, as is well known, dies out in a temperate climate, but it is liable to reappear on returning to the tropics. If it really were only the shafts of the hairs that were affected, it might be supposed that a rapid and efficient means of dealing with the condition would be shaving. This has been tried in some cases, but unless other means were adopted at the same time, the disease reappeared as soon as the hairs had re-grown to a length of half or three-quarters of an inch.

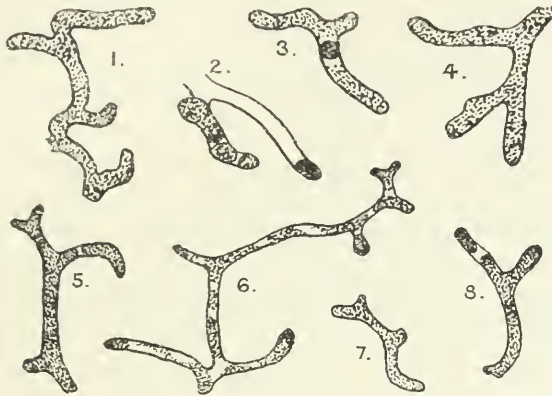
THE CAUSAL ORGANISM—*NOCARDIA TENUIS*

Scrapings from the infected hairs of all the cases I have examined have shown great numbers of hyphae, the majority of which were similar to those described by Chalmers and O'Farrell, namely, 'narrow, elongated, unbranched, non-septate rods resembling bacilli.' The hyphae usually stained deeply, but some only faintly, as though they were mere empty shells. Intensely stained granules were common, and occurred both at the ends and in the middle of the hyphae. The organism was Gram-positive and not acid-fast, and appeared to be identical with *Nocardia tenuis*, the causal agent of *Trichonocardiasis* described by Castellani (1911).

Chalmers and O'Farrell state that they did not meet with any branched forms in the nodules on the hairs, but they observed them in the slight growth they obtained in hanging drop cultures of blood serum and saline solution. Castellani and Chalmers (1913), however, state that the hyphae found in the nodules are 'occasionally branching.' In the cases I have examined in the Gold Coast, branched hyphae were always easily found (see figs. 1 to 4).

Measurements of such an organism are necessarily somewhat unsatisfactory owing to the diversity of forms found in the scrapings from the hairs. The lengths, 2μ to over 7μ , given by Chalmers and O'Farrell, and 4μ to 10μ given by Castellani and Chalmers, were

of course greatly exceeded in my cases in those hyphae which were branched. There seems to be some uncertainty about the breadth of the hyphae of *Nocardia tenuis*. According to Castellani and Chalmers (1913) 'they are rather thin, 1 to $1\frac{1}{2}\mu$,' but according to Chalmers and O'Farrell (1913) the average breadth is 'from $0\cdot14\mu$ to $0\cdot3\mu$.' The latter authors, however, figure (Plate XXXVI, figs. 10 and 12; Plate XXXVII, fig. 21) some hyphae which appear to measure $0\cdot6\mu$. The breadth of the hyphae in my cases was about $0\cdot6\mu$.



FIGS. 1 to 4.—Branched hyphae in scrapings from axillary hairs of a case of *Trichonocardiasis flava*. $\times 3000$.

FIGS. 5 to 8.—Branched hyphae from a culture a month old from the same hairs. $\times 3000$.

Nocardia tenuis has not previously been cultivated, but as has already been mentioned, Chalmers and O'Farrell observed some growth in hanging drops of equal parts of human blood serum and $0\cdot85$ per cent. normal saline solution.

Scrapings from the infected hairs of one of my cases of *Trichonocardiasis flava* were soaked in absolute alcohol and then planted on ascitic agar. After a few days' incubation at 37°C . a slight translucent growth was observed in one tube which on examination proved to be a pure culture of long, narrow hyphae. The colony grew slowly, and remained translucent and almost invisible. Sub-cultures on ascitic agar showed some growth within twenty-four hours, but the colonies were never luxuriant and spread

slowly over the surface of the medium as a thin transparent growth, the centre only having a slightly opaque ground-glass appearance. No difficulty was experienced in keeping the strain going on this medium, but sub-cultures on to ordinary agar were invariably unsuccessful.

The growth on ascitic agar at first consisted of bacillary bodies and long slender hyphae about 0.6μ in diameter. They were non-motile Gram-negative, and not acid-fast. The hyphae were unbranched, not septate, often of great length, and at the ends of some of the filaments a few short pieces appeared to be separated. After two days' growth the characters of the culture were similar, but the reaction, when stained by Gram's method, was less uniform, and some of the organisms retained the blue stain wholly or in part. The long hyphae were generally, if not always, unbranched. Successive examinations showed that as the culture aged the organism became more polymorphic and more Gram-positive, and after a week branched hyphae were easily found. Cultures a fortnight old were largely Gram-positive, and showed clubbed and branched hyphae of various sizes and shapes, but still of about the same diameter. Cultures a month old consisted of a mass of granular material in which short pieces of hyphae were distributed. These fragments were of all sizes, coccoid, bacillary, often branched, and very similar to the bodies found in the scrapings from infected hairs. They were Gram-positive, about 0.6μ broad, and the larger branched pieces showed the hypha in process of breaking up (see figs. 6 and 8). So closely did the organism in such a culture resemble that seen in smears made from scrapings of the hairs of the original case, that it could hardly be doubted but that they were the same.

On examining infected hairs stained by Gram's method a notable difference in reaction was observed. The larger nodules and those at the bases of the more heavily infected hairs retained the stain so that it was usually impossible to make out any details, but in the least dense portions Gram-positive organisms could sometimes be distinguished. The very young nodules and the areas of the hairs in which the infection was just commencing were, on the contrary, either completely or partially decolourised, and in specimens that had been counter-stained Gram-negative filaments

and bacillary bodies were seen in abundance. In a single hair, starting from its tip, all the stages observed in cultures on ascitic agar could be traced, from long Gram-negative filaments to short Gram-positive bodies of the type named by Castellani *Nocardia tenuis*. It must be concluded, therefore, that the characters of *Nocardia tenuis* as seen in the nodules on the hairs of a well-established case of trichonocardiasis represent only one phase in the life of this fungus, the nodules in fact corresponding to a culture of some considerable age.

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